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THE EFFECT OF WATER STRESS ON THE NITROGEN METABOLISM OF TWO MAIZE LINES:

1. EFFECTS ON THE PROTEIN CONTENT AND RNASE ACTIVITY

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ABSTRACT

The effect of drought conditions on the relative percentage water deficit, protein concentration and RNase activity was investigated in two maize lines. There seems to be a good correlation between protein content, RNase activity and drought resistance.

UITTREKSEL

DIE INVLOED VAN WATERSPANNING OP DIE STIKSTOFMETABOLISME VAN TWEE MIELIELYNE:
1. INVLOED OP DIE PROTEÏENINHOUD EN RNASE AKTIWITEIT

Die invloed van droogtetoestande op die relatiewe persentasie watertekort, proteïeninhoud en RNase aktiwiteit is by twee mielielyne ondersoek. Daar is blykbaar 'n noue verband tussen proteïeninhoud, RNase aktiwiteit en droogtebestandheid.

INTRODUCTION

Protein content of plants decreases during drought periods (Stutte & Todd, 1969; Savitskaya, 1972 and Dhindsa & Cleland, 1975). This is probably caused by the hydrolysis of proteins (Stocker, 1961) and an interruption of the protein synthesis system by a higher RNase activity (Arad & Richmond, 1976 and Maranville & Paulsen, 1972). There seems to be a good correlation between protein content and drought resistance. Drought-resistant plants have a higher protein content than plants with poorer drought resistance (Todd *et al.*, 1962). Estimation of protein content therefore might be an effective method for selection of drought resistance in plants (Stutte & Todd, 1967).

MATERIAL AND METHODS

Two maize lines, A281 and G556 DT, differing in drought resistance were used as experimental material. The plants were grown in controlled environmental cabinets in plastic pots of 150 mm diameter and with 3 kg of light red sandy loam as the root medium. Fertilizer (3:2:1) was added to the soil in the ratio of 1 g fertilizer to 1 kg of soil. The day length was 13 h, temperature 29 °C, the relative

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humidity 35% and the light intensity 14 530 lux. Night temperature was 20 °C and the relative humidity during the night 40%. After germination the plants were watered every second day with 200 mℓ water for 5 weeks. Thereafter only the control plants were watered as usual. Experimental samples were taken on the first, third, fifth and seventh day after the plants had last been watered. Ten plants of each maize line were cut off just above the ground and then cut into small pieces. Nine replicates of all the determinations were used.

Relative percentage water deficit (RWD) was estimated according to a method based on that of Stocker (1929). The RWD was calculated as follows:

$$\frac{\text{Turgor mass} - \text{Fresh mass}}{\text{Turgor mass} - \text{Dry mass}} \times \frac{100}{1}$$

Turgor mass of the tissue was obtained after the leaf segments had been floating on distilled water for 24 h.

Total water-soluble proteins were extracted according to a method based on that of Van Loon & Van Kammen (1968). One g of the leaf tissue was homogenised in 25 mℓ ice cold 0,1 M Tris (hydroxymethyl)-aminomethan buffer (pH 8,0). The extract was centrifuged for 15 minutes at 30 000 × gravity at a temperature between 0–2 °C. The supernatant was used for the determination of total water soluble proteins according to the method of Lowry, *et al.* (1951). Determinations on bovine serum albumin were used to plot a calibration curve.

The method used for the determination of RNase activity was based on the method used by Arad *et al.* (1973). One g of the leaf tissue was frozen in liquid air and then homogenised in 10 mℓ ice cold 0,1 M phosphate buffer (pH 6,0). The homogenate was centrifuged for 15 minutes at 15 000 × gravity at a temperature between 0–2 °C. One mℓ of the supernatant was added to 2 mℓ of substrate solution (yeast RNA 4 mg/mℓ in 0,1 M phosphate buffer pH 6,0) in test tubes. The assay mixture was incubated at 40 °C for 1 hour. Undigested RNA was precipitated with 1 mℓ of chilled 25% perchloric acid containing 0,75% uranylacetate. The tubes were left at 0–2 °C overnight and then centrifuged at 7 000 × gravity for 10 minutes. The absorbance of the supernatant was measured at 260 nm.

RESULTS

The results in Figure 1 show a linear increase in the RWD of both maize lines with increasing water stress. The RWD of A281, however, was higher than that of A281 throughout the stress period. Especially from the third day onwards there was evidently a more rapid desiccation of A281.

Estimation of the water soluble protein content showed that G556DT had a higher content than A281. The protein content of both maize lines decreased from the first day, and especially from the fifth day onwards there is a rapid decrease

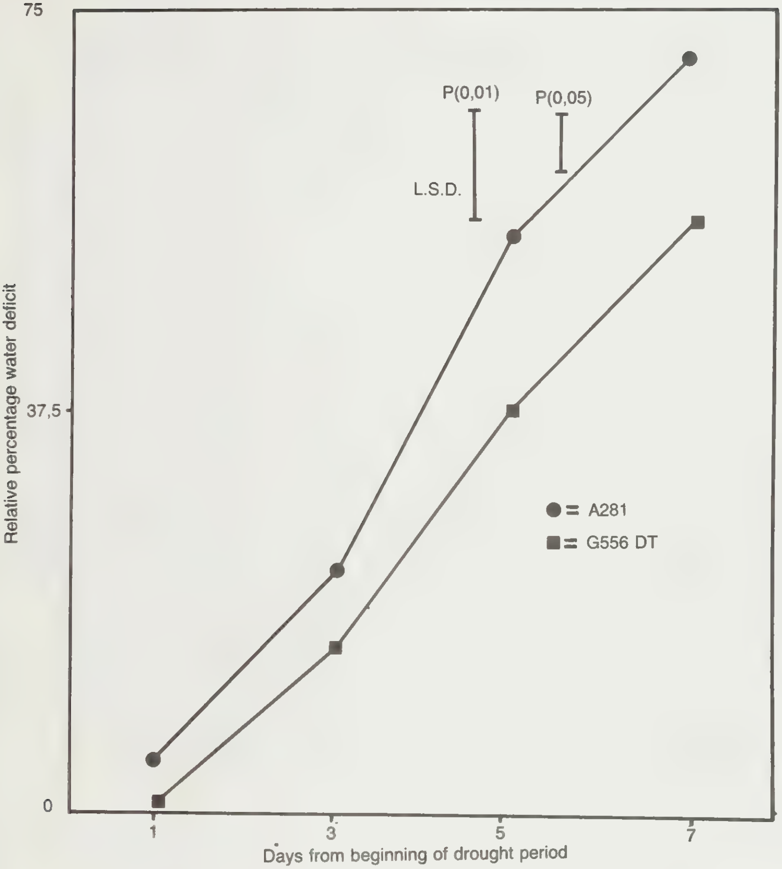

FIG. 1.
Changes in the relatative percentage water deficit of the two maize lines during a period of increasing water stress.

(Fig. 2). The percentage decrease, however, is much lower especially between day one and five, in G556DT than in A281 (Fig. 3).

A281 showed an increase in RNase activity from the first day of the drought period and a very high increase from the fifth day onwards. G556DT only showed an increase after the third day, and the RNase activity was lower in this line than that of A281 throughout the stress period (Fig. 4).

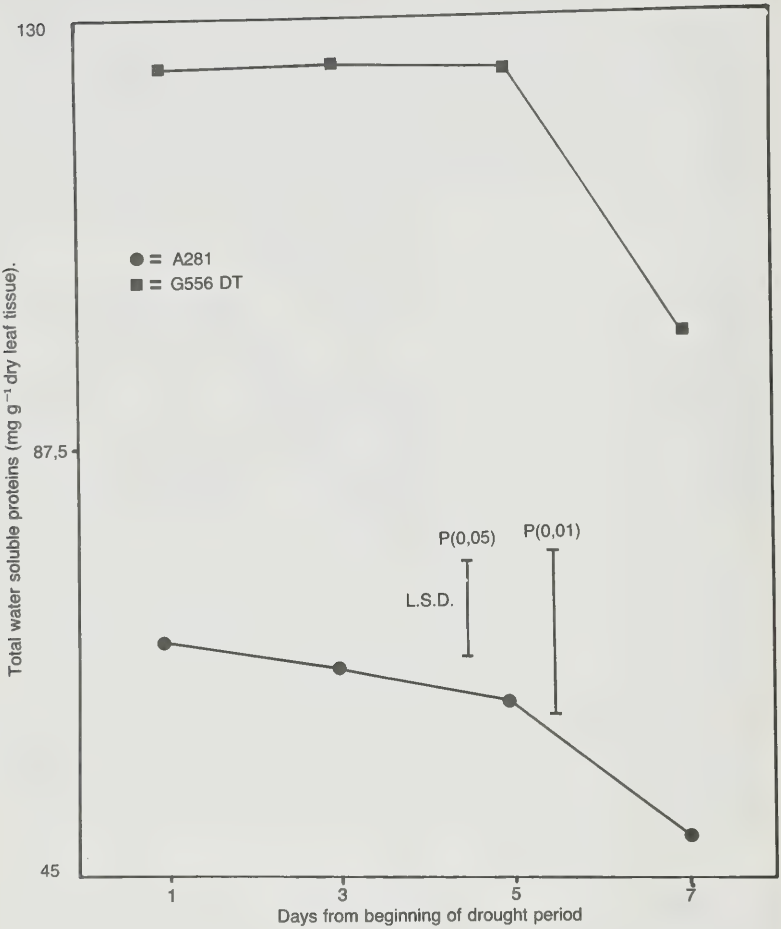

FIG. 2.
Changes in the total water soluble protein content (mgg^{-1} dry leaf tissue) of the two maize lines during the drought period.

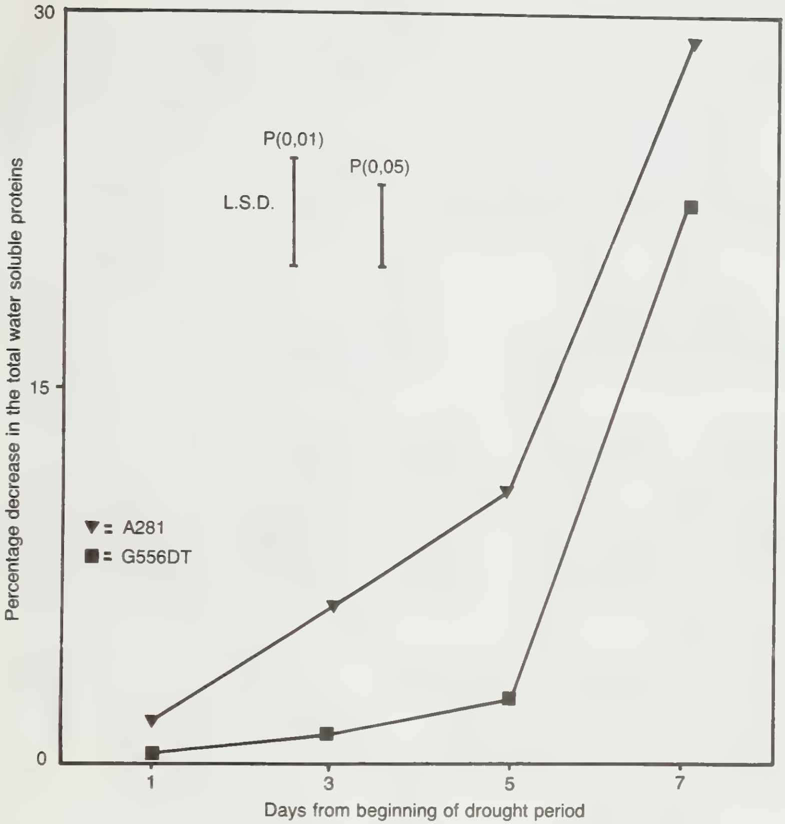

FIG. 3.
The percentage decrease in the total water soluble protein content during the drought period.

DISCUSSION

According to Chen *et al*. (1964) protein effects constitute the main component of the matrix potential of plant tissue. The higher water-soluble protein content of G556DT is probably the main reason why this maize line is able to delay the development of a water deficit in the tissue. Similar results to those reported in this communication were obtained by Todd *et al*. (1962), who found that wheat with high drought resistance had a greater protein content than wheat with a lower drought resistance.

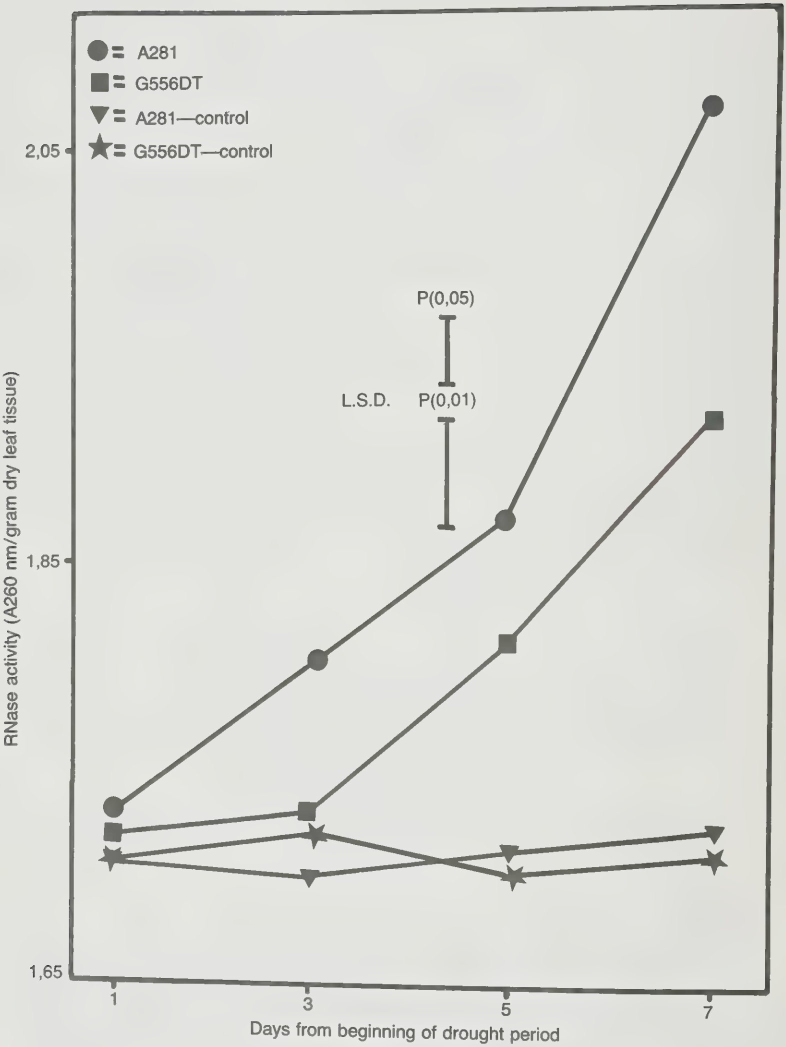

FIG. 4.
Changes in the RNase activity (A260 nm/gram dry leaf tissue) of the two maize lines during the drought period.

Conditions of water stress cause a decline in the protein content of both maize lines. Dhindsa & Bewley (1976), Maranville & Paulsen (1972) and Savitskaya (1972) also observed such decreases in protein content during drought conditions. It is, however, apparent that the decrease in protein content is smaller in G556DT than in A281, especially between days one and five (Fig. 3). This sharp decline in A281, is probably caused by the hydrolysis of proteins and a partial interruption of the protein synthesis system. Dehydration causes breakdown of the secondary and tertiary structure of proteins and this leads to an easier hydrolysis of the proteins (Chen *et al.*, 1964).

One of the main factors affecting protein synthesis is RNase activity. The RNase activity evidently affects the rate of protein synthesis by destroying the mRNA linking the ribosomes (Henckel, 1970). In this experiment the increase in RNase activity is much higher in A281 than in G556DT throughout the water stress period, providing a possible explanation for the protein content differential between the two lines.

From the water deficit data it is evident that G556DT is more drought resistant than A281. This might be due to the higher protein content caused by the lower RNase activity during increasing water stress and possibly also by a more stable protein structure in the former maize line. Since the protein content of G556DT remained much higher than that of A281 throughout the periods of water stress, the results of this investigation support the idea that estimation of protein content may be a method for the selection of drought-resistant maize lines.

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